

Amendments to the Claims:

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

Please cancel original claims 1-41 and add the following new claims 42-81:

42. (New) A method for producing a nucleic acid product, comprising that an isolated polymerase protein is contacted with a nucleic acid template under conditions sufficient for the function of the enzyme, and wherein said polymerase protein is an RNA polymerase capable of producing
- (a) short complementary RNA copies of said template, which are scattered throughout the entire template length and, optionally,
- (b) template-length complementary RNA copies.
43. (New) The method according to claim 42, wherein said nucleic acid template is DNA or RNA.
44. (New) The method according to claim 42, wherein the ratio of said short and template-length RNA copies can be adjusted by the reaction conditions.
45. (New) The method according to claim 42, wherein the length of said short RNA copies can be adjusted by the reaction conditions.
46. (New) The method according to claim 42, wherein the said short or template-length RNA copies are annealed to the template or denatured from the template.
47. (New) The method according to claim 42, wherein said nucleic acid template is linear or circular.
48. (New) The method according to claim 42, wherein said polymerase originates from a eukaryotic cell.
49. (New) The method according to claim 48, wherein said polymerase originates from an organism selected from the kingdoms of Fungi, Viridiplantae, Metazoa, or the group of Mycetozoa.
50. (New) The method according to claim 48, wherein said polymerase originates from an organism selected from the subset of genera *Neurospora*, *Arabidopsis*, *Caenorhabditis*, and *Dictyostelium*,

preferably organisms *Neurospora crassa*, *Arabidopsis thaliana*, *Caenorhabditis elegans*, and *Dictyostelium discoideum*.

51. (New) The method according to claim 50, wherein said polymerase is QDE-1 protein of *Neurospora crassa* or an altered or a genetically modified derivative of QDE-1.

52. (New) The method according to claim 42, wherein said RNA polymerase is encoded by a nucleic acid sequence selected from the group of:

- (a) a nucleic acid sequence comprising the nucleic acid sequence of SEQ ID NO: 1 or SEQ ID NO: 3;
- (b) a nucleic acid sequence encoding a polypeptide comprising the amino acid sequence of SEQ ID NO 2 or SEQ ID NO: 4;
- (c) a nucleic acid sequence, which differs from the nucleic acid sequence of (a) or (b) due to degeneracy of the genetic code;
- (d) a nucleic acid sequence hybridizing to the nucleic acid sequence of (a), (b) and/or (c) under stringent conditions; and
- (e) a nucleic acid sequence encoding a polypeptide comprising the amino acids 709 to 1402 of SEQ ID NO:4 or any sequence longer than that up to the sequence comprising the amino acids 2 to 1402 of SEQ ID NO: 2; and
- (f) a nucleic acid sequence encoding an amino acid sequence, which shows at least 50% identity to the amino acid sequence of SEQ ID NO: 2.

53. (New) The method according to claim 42, comprising the steps of:

- (a) providing ssRNA or ssDNA template;
- (b) contacting said ssRNA or ssDNA template *in vitro* with the polymerase under conditions sufficient for RNA synthesis.

54. (New) The method according to claim 53, wherein said ssRNA template is provided by transcribing a DNA template with a DNA-dependent RNA polymerase, preferably polymerase selected from the

group of DNA bacteriophage-encoded DNA-dependent RNA polymerases, most preferably DNA-dependent RNA polymerase of bacteriophage T7, T3 or SP6.

55. (New) The method according to claim 53, wherein the reactions are carried out at the same time or sequentially in the same reaction vessel.

56. (New) The method according to claim 53, wherein the newly produced RNA species are recovered from the reaction mixture.

57. (New) The method according to claim 53, wherein said newly produced RNA strands are annealed with the template to form dsRNA elements or, alternatively, are denatured from the template.

58. (New) The method according to claim 53, wherein RNA synthesis is initiated from the 3' end of a nucleic acid primer complementary to the RNA or DNA template or RNA synthesis is initiated without a primer.

59. (New) The method according to claim 53, wherein the reaction mixture for RNA synthesis comprises at least one nucleoside triphosphate optionally labeled with a radioactive isotope or is chemically modified, pH buffer, ammonium acetate, PEG, Mg<sup>2+</sup> ions, Mn<sup>2+</sup> ions and/or non-ionic detergent.

60. (New) The method according to claim 59, wherein the method is specifically used for producing radioactively or chemically labeled RNA probes and comprising an optional step of purifying the newly produced labeled RNA from the components of the reaction mixture.

61. (New) The method according to claim 60, wherein said labeled RNAs are used as probes for Southern or Northern blot analyses after the optional purification step.

62. (New) The method according to claim 60, wherein said labeled RNAs are used as probes for a fluorescent *in situ* hybridization analysis after the optional purification step.

63. (New) The method according to claim 60, wherein said labeled RNAs are used as probes for a microarray analysis after the optional purification step.

64. (New) An isolated polypeptide, characterized in that:

(i) said polypeptide has sufficient RNA polymerase activity and is capable, when contacted with a nucleic acid template, of producing short complementary RNA copies of said template, which RNA copies are scattered throughout the entire template length and, optionally, capable of producing template-length complementary RNA copies;

(ii) said polypeptide has enhanced solubility resulting in at least 3 times higher yield of the active polymerase, than in the case of polypeptide comprising the amino acid sequence of SEQ ID NO: 2 or encoded by the nucleic acid sequence comprising SEQ ID NO: 1; and

(iii) said polypeptide is encoded by a nucleic acid sequence selected from the group of:

(a) a nucleic acid sequence comprising the nucleic acid sequence of SEQ ID NO: 3;

(b) a nucleic acid sequence encoding a polypeptide comprising the amino acid sequence of SEQ ID NO: 4;

(c) a nucleic acid sequence, which differs from the nucleic acid sequence of (a) or (b) due to degeneracy of the genetic code;

(d) a nucleic acid sequence hybridizing to the nucleic acid sequence of (a), (b) and/or (c) under stringent conditions;

(e) a nucleic acid sequence encoding a polypeptide comprising the amino acids 709 to 1402 of SEQ ID NO:4 or any sequence longer than that up to the sequence comprising the amino acids 2 to 1402 of SEQ ID NO: 2; and

(f) a nucleic acid sequence encoding an amino acid sequence, which shows at least 50% identity to the amino acid sequence SEQ ID NO: 2.

65. (New) An isolated nucleic acid sequence encoding the polypeptide according to claim 64.

66. (New) A vector, which comprises the nucleic acid sequence of claim 65 operationally linked with regulatory sequences required for gene expression.

67. (New) A host cell comprising the vector of claim 66.

68. (New) A method for producing a polymerase protein, which comprises culturing the host cell of claim 67 under conditions suitable for the expression of the protein.

69. (New) The method according to claim 68, comprising the step that the protein is recovered from the cell or culture medium and optionally purified.

70. (New) A method for studying nucleic acid secondary structure, preferably RNA secondary structure comprising the steps of:

- (a) providing nucleic acid target molecule, preferably RNA target molecule;
- (b) contacting said target molecule with an isolated RNA polymerase capable of producing short complementary RNA copies of the nucleic acid target as template, which RNA copies are scattered throughout the ssRNA regions of the template length and, optionally, template-length complementary RNA copies under conditions sufficient for RNA synthesis in a mixture additionally comprising radioactively or chemically labeled nucleotides, so that single-stranded elements of said target RNA are copied by the polymerase;
- (c) recovering and optionally purifying the newly produced labeled nucleic acid species from the reaction mixture;
- (d) using said labeled nucleic acid species as probes for microarray chip that comprises nucleic acid fragments of said target molecule;
- (e) interpreting data from the microarray analysis to deduce which parts of the target molecule are single-stranded; and optionally
- (f) building a model for the secondary or tertiary structure of the target molecule.

71. (New) A method for studying nucleic acid-protein interactions, preferably RNA-protein interactions comprising the steps of:

- (a) providing a nucleic acid target and nucleic acid binding protein, preferably an RNA target and an RNA-binding protein;

(b) contacting said target and the solution of said protein in an experimental mixture under conditions sufficient for target-protein interaction, and in a separate vessel, contacting said target with a control solution that lacks said protein.

(c) contacting said experimental and control mixtures with an isolated RNA polymerase capable of producing short complementary RNA copies of said target as template, which RNA copies are scattered throughout the template length not

covered by protein and, optionally, template-length complementary RNA copies under conditions sufficient for RNA synthesis;

(d) recovering and optionally purifying the newly produced labeled nucleic acid species from both reaction mixtures;

(e) using the two sets of labeled nucleic acid species as probes for two identical microarray chips that comprise nucleic acid fragments of the target;

(f) interpreting data from the two microarray analyses to deduce which parts of the target molecule are accessible for the RNA synthesis;

(g) comparing the two data sets to determine the difference between target in experimental and control mixtures; and optionally

(h) interpreting the difference between the two data sets as a model for nucleic acid-protein interactions

72. (New) A method for producing RNA trigger molecules to induce RNA interference *in vivo* or *in vitro*, comprising the steps of:

(a) providing RNA or DNA template;

(b) contacting said RNA or DNA template with an isolated RNA polymerase capable of producing short complementary RNA copies of said template, which are scattered throughout the entire template length and, optionally, template-length complementary RNA copies under conditions sufficient for RNA synthesis in a mixture comprising: nucleic acid template, said RNA polymerase, nucleoside triphosphates, and optionally pH buffer, ammonium acetate, PEG, Mg<sup>2+</sup> ions, Mn<sup>2+</sup> ions and/or non-ionic detergent; and

(c) incubating the reaction mixture at temperature sufficient for RNA synthesis.

73. (New) The method according to claim 72, wherein said RNA or DNA template originates from a cell or a virus.

74. (New) The method according to claim 72, wherein said RNA template is provided by transcribing a DNA template with a DNA-dependent RNA polymerase, preferably derived from a bacteriophage selected from the group of T7, T3, and SP6 bacteriophages.

75. (New) The method according to claim 72, wherein steps (a) and (b) are carried out at the same time or sequentially in the same reaction vessel.

76. (New) A kit comprising the polypeptide of claim 64.

77. (New) The kit according to claim 76, wherein the kit further comprises additives necessary for a detectable level of RNA synthesis.

78. (New) The kit according to claim 76 comprising nucleoside triphosphates in concentrations sufficient for RNA synthesis.

79. (New) The kit according to any claim 76, wherein at least one nucleoside triphosphate is labeled with a radioactive isotope or is chemically modified.

80. (New) The kit according to claim 76, additionally comprising a standard nucleic acid preparation (or preparations) with characterized capacity to serve as a template (templates) for RNA synthesis by the polypeptide characterized in that:

(i) said polypeptide has sufficient RNA polymerase activity and is capable, when contacted with a nucleic acid template, of producing short complementary RNA copies of said template, which RNA copies are scattered throughout the entire template length and, optionally, capable of producing template-length complementary RNA copies;

(ii) said polypeptide has enhanced solubility resulting in at least 3 times higher yield of the active polymerase, than in the case of polypeptide comprising the amino acid sequence of SEQ ID NO: 2 or encoded by the nucleic acid sequence comprising SEQ ID NO: 1; and

- (iii) said polypeptide is encoded by a nucleic acid sequence selected from the group of:
- (a) a nucleic acid sequence comprising the nucleic acid sequence of SEQ ID NO: 3;
  - (b) a nucleic acid sequence encoding a polypeptide comprising the amino acid sequence of SEQ ID NO: 4;
  - (c) a nucleic acid sequence, which differs from the nucleic acid sequence of (a) or (b) due to degeneracy of the genetic code;
  - (d) a nucleic acid sequence hybridizing to the nucleic acid sequence of (a), (b) and/or (c) under stringent conditions;
  - (e) a nucleic acid sequence encoding a polypeptide comprising the amino acids 709 to 1402 of SEQ ID NO:4 or any sequence longer than that up to the sequence comprising the amino acids 2 to 1402 of SEQ ID NO: 2; and
  - (f) a nucleic acid sequence encoding an amino acid sequence, which shows at least 50% identity to the amino acid sequence SEQ ID NO: 2.

81. (New) Use of the method of claim 42 for studying nucleic acid secondary structure, for studying nucleic acid-protein interactions, or for producing RNA trigger molecules.